

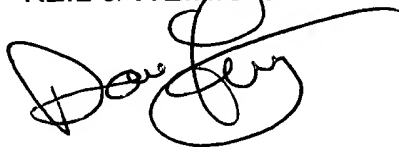
REMARKS

Further in response to the examiner's office action of January 4, 2002, and additionally in response to the examiner's office communication of August 26, 2002, applicants amend claims 12 and 22 herein. These amendments remove certain language concerning the concept of "substrate specificity," and should be sufficient to overcome the rejection under 35 USC §112, ¶2.

In view of the foregoing amendments and remarks, applicants consider that the rejections of record have been obviated and respectfully solicit passage of the application to issue.

Please charge any shortage in fees due in connection with the filing of this paper, including Extension of Time fees to Deposit Account No. 11-0345. Please credit any excess fees to such deposit account.

Respectfully submitted,  
KEIL & WEINKAUF

A handwritten signature in black ink, appearing to read "David C. Liechty", with a large, stylized flourish extending from the end of the signature.

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**VERSION WITH MARKINGS TO SHOW CHANGES MADE**

**IN THE CLAIMS**

Please amend claims 12 and 22 to read as follows:

12. (amended) A method for altering the substrate specificity of an enzyme [to a substrate from a substrate specificity where catalysis does not occur to a substrate specificity where catalysis does occur], comprising the steps of:
- a) introducing a DNA sequence coding for the enzyme into the *Escherichia coli* strain XL1-Red or into a functional derivative thereof,
  - b) incubating the transformed *Escherichia coli* strain XL1-Red or its functional derivative to generate mutations in the DNA sequence,
  - c) transferring the mutated DNA sequence from the transformed *Escherichia coli* strain XL1-Red or its functional derivative to a microorganism which has no impeding enzyme activity,
  - d) incubating this microorganism to detect the enzyme activity in at least one selection medium which comprises at least one enzyme substrate to recognize altered substrate specificity of the enzyme, with or without other indicator substances,
  - e) selecting the microorganisms which show an alteration in the substrate specificity, said microorganisms in steps b), d) and e) being a member selected from the group consisting of bacteria, fungi and yeasts,
- wherein the enzyme is selected from the group consisting of lipases, amidases,

nitrilases, ether hydrolases, peroxidases, glycosidases and phytases.

22. (amended) A method for altering the substrate specificity of an enzyme [to a substrate from a substrate specificity where catalysis does not occur to a substrate specificity where catalysis does occur], comprising the steps of:
- a) introducing a DNA sequence coding for the enzyme into the *Escherichia coli* strain XL1-Red or into a functional derivative thereof,
  - b) incubating the transformed *Escherichia coli* strain XL1-Red or its functional derivative to generate mutations in the DNA sequence,
  - c) transferring the mutated DNA sequence from the transformed *Escherichia coli* strain XL1-Red or its functional derivative to a microorganism which has no impeding enzyme activity,
  - d) incubating this microorganism to detect the enzyme activity in at least one selection medium which comprises at least one enzyme substrate to recognize altered substrate specificity of the enzyme, with or without other indicator substances,
  - e) selecting the microorganisms which show an alteration in the substrate specificity, said microorganisms in steps b), d) and e) being a member selected from the group consisting of bacteria, fungi and yeasts,
- wherein the enzyme is an esterase selected from the group consisting of *Pseudomonas fluorescens* esterase, pig liver esterase and *Thermoanaerobium brockii* esterase.